Supplemental Materials and Methods

iChIP using r3xFNLDD-D with Dock Catch Resin

Cells (2×10^7) were fixed with 1% formaldehyde at 37°C for 5 min. The chromatin fraction was extracted and fragmented (2 kbp-long on average) by sonication as described previously [12] except for using 800 μl of Calcium Buffer (10 mM Tris pH 8.0, 150 mM NaCl, 2 mM $CaCl_2$) and Ultrasonic disruptor UD-201 (TOMY SEIKO). After sonication, TritonX-100 was added to final concentration at 0.1%. The sonicated chromatin (400 μl) was pre-cleared with 30 μl of mouse IgG-agarose (Sigma-Aldrich) and subsequently incubated with 30 μl of Dock Catch Resin (Sysmex Corporation) at 4°C for 20 h. Dock Catch Resin was washed four times with 1 ml of Calcium Buffer with 0.1% TritonX-100 and once with 1 ml of Calcium Buffer with 0.1% IGEPAL-CA630. The isolated chromatin complexes were eluted with 120 μl of Elution Buffer (50 mM Tris pH 7.5, 150 mM NaCl, 5 mM EGTA, 0.1% IGEPAL-CA630) at 25°C for 30 min. After reverse crosslinking at 65°C, DNA was purified with ChIP DNA Clean & Concentrator (Zymo Research) and used as template for real-time PCR with SYBR Select PCR system (Applied Biosystems) using the Applied Biosystems 7900HT Fast Real-Time PCR System.

RT-PCR

Total RNA extracted with Isogen II (Nippon gene) was used as template for reverse transcription with ReverTra Ace qPCR RT Master Mix with gDNA Remover (Toyobo). The cDNA was used as template for PCR with AmpliTaq Gold 360 Master Mix (Applied Biosystems). PCR cycles were as follows: heating at 95°C for 10 min; 30 - 35 cycles of 95°C for 30 sec, 60°C for 30 sec,

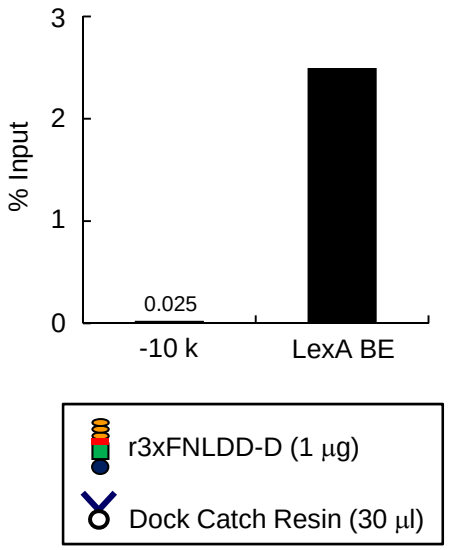
72°C for 1 min; and the final extending 72°C for 2 min. The primers used in this experiment are shown in Table 1.

Supplemental Figure Legends

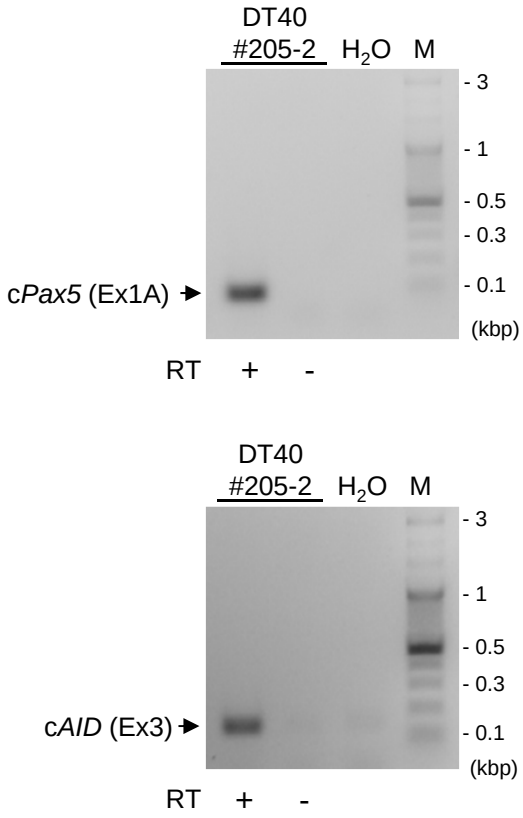
Supplemental Figure S1. iChIP using r3xFNLDD-D with Dock Catch Resin.

Supplemental Figure S2. RT-PCR analysis of c*Pax5* and c*AID* mRNA. Total RNA was extracted from DT40#205-2 and used in RT-PCR for detection of c*Pax5* mRNA (upper panel) and c*AID* mRNA (lower panel). M: Molecular size markers.

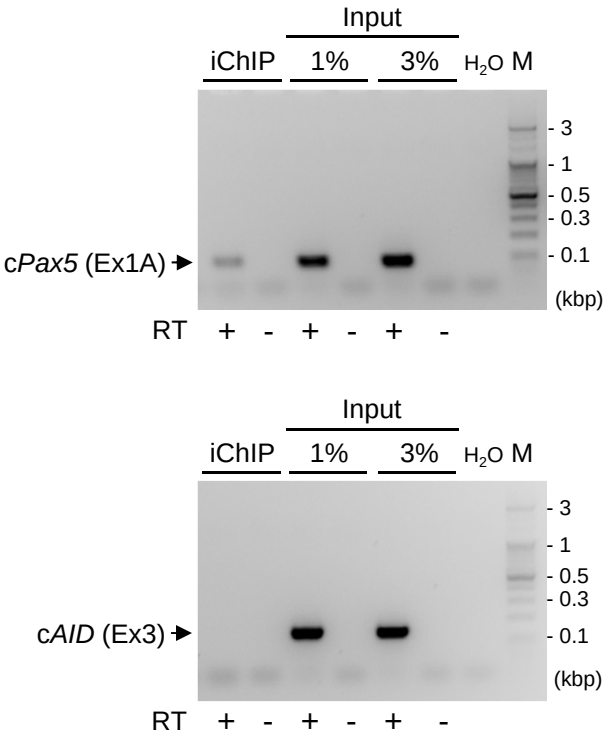
Supplemental Figure S3. The full-length images of Figure 5C including molecular size markers. c*Pax5* (Ex1A) and c*AID* (Ex3) are shown in the upper panel and lower panels, respectively. M: Molecular size markers.



Supplemental Figure 1. Fujita and Fujii



Supplemental Figure 2. Fujita and Fujii



Supplemental Figure 3. Fujita and Fujii